



Efficiency of a hybrid solid digestion-denitrification column in suspended solid and nitrate removal from recirculating aquaculture system

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ABSTRACT

This research focused on the solid and nitrate removal efficiency in a solid digestion-denitrification column. The 20 L up-flow column consisted of 18 L acrylic column with 2 L down-comer inlet tube located in the middle. In the first part, the wastewater with high suspended solids from the Tilapia fish tank was applied into the sedimentation unit at 5 variable flow rates i.e., 11.25, 25.71, 60, 105.88 and 360 L/h. The results indicated that the flow rate of 11.25 L/h (0.57 m/h) gave the highest solid removal efficiency of 72.72% ± 8.24%. However, the total suspended solids removal was highest at 360 L/h (18.13 m/h). In the second part, methanol was added as an external organic carbon source for denitrification process in a hybrid column containing settled solids. The COD:N ratios of 0.5:1, 1:1, 2:1, 3:1, 4:1, 5:1 and 6:1 were investigated and compared with control without methanol addition. This experiment was operated at the HRT of 1 h with 450 L wastewater from recirculating aquaculture pond containing 100 mg-N/L sodium nitrate. The results indicated that the COD:N ratio of 3:1 gave the highest nitrate removal efficiency of 33.32% ± 21.18% with the denitrification rate of 5,102.88 mg-N/day.

Keywords: Denitrification, Hybrid column, Nitrate, Recirculating aquaculture system, Sedimentation, Solid digestion

1. Introduction

Recently, aquaculture production has grown rapidly as a response to the demand of good quality protein food. Intensive aquaculture using the recirculating aquaculture system (RAS) is therefore the promising manner of the future for sustainable food production. With the RAS, fishes were grown at high density to produce large quantities in limited space. In addition, RAS reduces water consumption and waste discharge in land-based aquaculture where as the appropriate technologies are required to maintain good water quality in the pond [1]. However, the major problems of RAS are the gathering of suspended solids derived from fecal, uneaten feed and the accumulation of inorganic nitrogen compounds such as ammonia, nitrite and nitrate in the water [2]. Suspended solid can deposit at the bottom of the pond and decomposed by either aerobic or anaerobic microorganisms. As a result, hydrogen sulfide produced from the anaerobic sulfate reduction can harm the aquatic animals and even cause the mass mortality. Thus, it is necessary

to control suspended solid within the optimum concentration [3]. The successful recommended ways to remove excess suspended solids from fish tank can be classified into three techniques which are gravity separation, filtration and flotation [4].

In the aquaculture pond, the accumulation of inorganic nitrogen compounds as ammonia, nitrite and nitrate is the results from the biological degradation of animal excretion and the uneaten feed. Nitrogen wastes have a deleterious impact on water quality and fish growth [5]. Typically, RAS must retain ammonia concentration less than 0.025 mg-N/L by nitrification process in which ammonia is oxidized to nitrite and nitrate, respectively. Likewise, accumulation of nitrate in the RAS to the concentration higher than 50 mg-N/L may result in stress of aquatic animal and decrease food intake [6]. Therefore, reducing nitrate to nitrogen gas by denitrification process is necessary for long-term water recycle [7]. The effectiveness of denitrification process depends on various factors such as temperature, pH, dissolved oxygen (DO), nitrite concentration [8], and also the amount of organic carbon substrate that available in the system [9, 10]. Among these factors, many



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previous reports were specified that organic carbon substrate is the major significant in aquaculture denitrification process [11-14].

In this research, we aim to develop the alternative approach to remove the suspended solids from indoor recirculating aquaculture pond by simple gravity sedimentation process. Since sedimentation generally requires low energy input and easy to operate. Moreover, instead of discarding the organic waste solids that remaining in the up-flow sedimentation column, we tried to degrade those particulate organic matters through anaerobic decomposition. The product yielded can be used as the sole carbon source and electron donors for biological denitrification without further need for the extra carbon supplementation as normally required in the aquaculture system [15, 16]. The treated water will have lower concentration of nitrate, in which can be safely recycled back to the aquaculture pond.

2. Materials and Methods

The experimental system comprised of 3 equipment parts as (1) a fish tank, (2) an equalization tank and (3) a hybrid solid digestion-denitrification column as indicated in Fig. 1. The fish tank was a plastic container of 3.5 m in diameter and the total volume was 3,000 L. The Tilapia, *Oreochromis niloticus*, was cultivated at high density (17 kg/m³). The fishes were fed with an artificial feed at 2% of the total body weight per day. The system was operated in a completely closed system without water discharge. The equalization tank was a 500 L plastic tank with the working volume of 430 L. Wastewater from Tilapia tank was pumped into the equalization tank then mixed well by air bubble and aquarium pump in order to prevent suspended solid sedimentation and sludge decomposition. The hybrid solid digestion-denitrification column was an up-flow suspended solid separation unit which made from a transparency acrylic tube with the working volume of 20 L. The unit consisted of 2 layers of acrylic tubes. The outer tube was 17 cm in diameter and 100 cm height. The inner tube (down-comer tube) diameter was 5.4 cm with shorter height of 97 cm. Water from the equalization tank was pumped into the inner tube. The water flowed down to the bottom of the column and rose up in the outer tube before returned back to the equalization tank. Thus the heavy solids were accumulated at the bottom of the inner separation unit.

2.1. The Effect of Flow Rate on Solid Removal

As shown in Fig. 1, the hybrid column was connected with the equalization tank. In this experiment, this column reactor will perform as a solid separation unit. The water from the fish pond was pumped into the equalization tank and the water from the equalization was moved to the separation unit by a submersible pump and then returned back into the equalization tank. The flow rate of the water pass through the solid separation unit was set up and maintained by 3 ball valves. With the settling velocity of suspended solids from fish pond is about 2.55 m/h⁵, in this process, the flow rate was therefore varied with 5 levels including: 11.25, 25.71, 60, 105.88 and 360 L/h. These flow rates provided the different upflow velocity in the sedimentation column as 0.57, 1.29, 3.02, 5.33 and 18.13 m/h, respectively. The system was operated for 2 days for each flow rate. The water influent and effluent

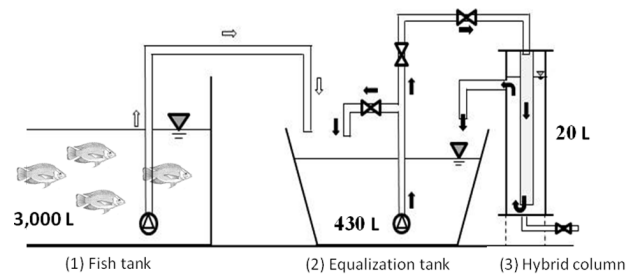


Fig. 1. The experimental system for the suspended solid separation.

were collected 10 times/d as every hour interval for suspended solid analysis according to the standard method [17].

2.2. The Optimal Conditions for Denitrification Process

The similar procedures were performed as in the first experiment; wastewater with high suspended solids from the fish pond was pumped into the equalization tank. Sodium nitrate was added in this tank then adjusts to the final concentration of 100 mg-N/L in a total volume of 430 L. The well-mixed wastewater was transferred to the column reactor by a submersible pump with the optimal flow rate of the up-flow sedimentation column from the previous experiment. The experiment was operated with the optimum hydraulic retention time in a reactor by water from the equalization tank. Methanol was added as an external organic carbon source using a peristaltic pump. The oxidation-reduction potential (ORP) probe was used to measure and control the ORP in the reactor not less than -150 mV (Fig. 2). The 7 variable of COD:N ratios as 0.5:1, 1:1, 2:1, 3:1, 4:1, 5:1 and 6:1 were investigated and compare with a non-methanol addition system. The influent and effluent water were collected everyday for the water quality analysis (ammonia, nitrite, nitrate, alkalinity, pH, dissolved oxygen and temperature) according to the standard method [17]. Ammonia and nitrite concentrations were analyzed using a colorimetric method, while the nitrate concentration was measured with a spectrophotometric screening method according to APHA, 1992 [18]. Alkalinity was measured using a test kit (Aquatic Animal Medicine Division, Department of Veterinary Medicine, Chulalongkorn University, Thailand) and residual methanol in the water from the denitrification tank on the last day was analyzed by gas chromatography (GC-2010; Shimadzu, Japan). Other parameters, such as pH, dissolved oxygen and temperature were measured using a pH meter (HI98240; Hanna Instruments, USA), DO meter (HI964400; Hanna Instruments, USA) and thermometer, respectively.

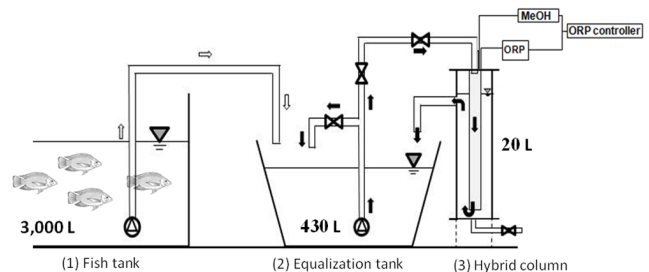


Fig. 2. The experimental system for denitrification activity determination.

3. Results and Discussion

3.1. The Effect of Flow Rate on Solid Removal

The solid removal efficiencies of the 2-days experiment for 5 upflow velocities at 0.57, 1.29, 3.02, 5.33 and 18.13 m/h are illustrated in Fig. 3. It was found that the slowest upflow velocity of 0.57 m/h (11.25 L/h) gave the highest solid removal efficiency of $72.72\% \pm 8.24\%$ (Fig. 4). With low surface loading rate, small particles could be gravitated according to the Stokes' law. The settling velocity was controlled by diameter of the particle, dynamic viscosity, density of water and density of the particle. The result was comparable with the previous research by Nurit [19] that the higher efficiency of solid separation was occurred at the lower flow rate.

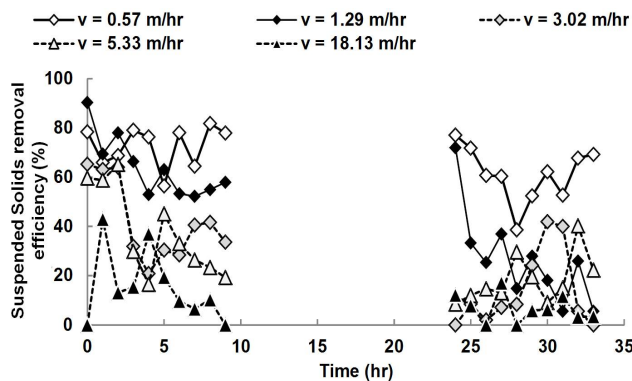


Fig. 3. The solids removal efficiency of 5 variable upflow velocities for a 2 days operated system.

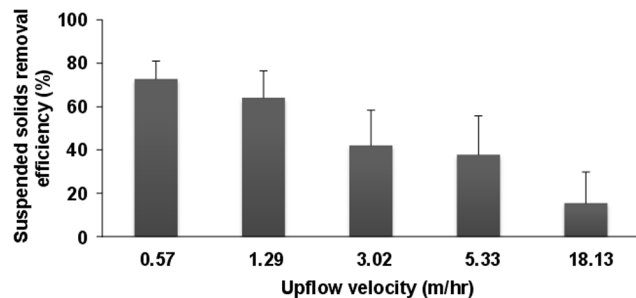


Fig. 4. The average of solids removal efficiency during the first 10 hours at each upflow velocity.

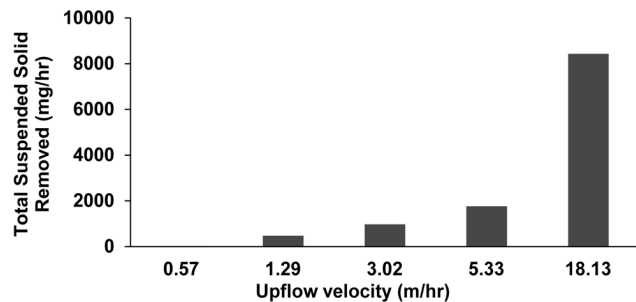


Fig. 5. The average of total suspended solid (TSS) removal rate during the first 10 hours at each upflow velocity.

However, when considering the solid removal rate, it can be seen that the upflow velocity of 18.13 m/h (360 L/h) gave the highest total suspended solid (TSS) removal up to 8,439.84 mg/h (Fig. 5). The flow rate of 11.25 L/h, on the other hand, gave the highest efficiency but the total solids removed were only 26.93 mg/h. As a result, the upflow velocity of 18.13 m/h was therefore selected and applied for further studies in order to develop the solid separation device for aquaculture propose.

3.2. The Optimal Conditions for Denitrification Process

The effect of methanol addition and the COD:N ratio on nitrate removal efficiency by denitrification process in a hybrid solid digestion-denitrification column was evaluated. In this experiment, the up-flow velocity as 1.01 of m/h was controlled in order to prolong the hydraulic retention time of aquaculture wastewater in a reactor to about 1 hour. The HRT period of 3.33 minutes from the upflow velocity of 18.13 m/h was too short to sustain the biological digestion process. Moreover, this duration was not enough for denitrifiers to achieve the complete denitrification activity in the solid digestion column. The result in Fig. 6 indicated the alteration of nitrate concentration in the influent and effluent at each COD:N ratio. It was found that the nitrate concentration of both influent and effluent from a control reactor (a non-methanol addition system) was quite stable during the whole experiment. There was less different in nitrate concentration between the inlet and outlet water, which referred as no nitrate removal in this column reactor. The average nitrate concentration in the water inlet and outlet were constant as 92.56 ± 3.88 and 93.08 ± 3.80 mg-N/L, respectively. While in the test systems with the COD:N ratios of 0.5:1, 1:1, 2:1, 3:1, 4:1, 5:1 and 6:1, the largely diverse in nitrate concentration of the influent and effluent were found. At the lower COD:N ratios of 0.5:1, the system required a longest period of 19 days to diminish the nitrogen concentration in the effluent to the safety level for aquaculture (below 30 mg-N/L). Whereas, this duration became shorter in the higher COD:N ratios which were 9 days, 9 days, 6 days, 6 days and 4 days of the COD:N ratios as 1:1, 2:1, 3:1, 4:1 and 5:1, with the shortest of 3 days for the ratio as 6:1. The results clearly implied that the addition of methanol in a hybrid solid digestion-denitrification column significantly accelerated the denitrification process, although the rate of nitrate removal depends on the amount of methanol added.

The results in Table 1 referred the diverse denitrification rates, nitrate removal efficiencies and the COD depletion among the 8 experimental conditions performed. It suggested that the digestion column with the COD:N ratios of 6:1 gave the maximum denitrification rate (9,182.88 mg-N/day), while the lowest denitrification (70.27 mg-N/day) was found in the control system without methanol addition. In this reactor, the nitrate removal efficiency was only $0.55\% \pm 1.17\%$. The highest nitrate removal efficiency of $33.32\% \pm 21.18\%$ was illustrated in the reactor with the COD:N ratios of 3:1. It was slightly higher than $31.44\% \pm 18.44\%$ in the reactor with the COD:N ratios of 6:1. The results emphasized the significant of external carbon source addition for nitrate removal activity in a solid digester column. Furthermore, the result was comparable with the previous research by Sutthisee

[20] which found that the higher efficiency of nitrate removal was occurred after the adequate addition of methanol. If the carbon source was not sufficient, the intermediate compounds of the reaction such as nitrite and nitrous oxide will take place due to the incomplete denitrification. In the opposite way, if methanol was added in the excess amount, not only the high cost of the system operation, the toxic effect will interfere the living of aquatic animals. In general, excess methanol must be avoided in an aqua-

culture system. Kaviraj *et al.* [21] reported that a methanol concentration greater than 47.49 mg/L (equivalent to 71.23 mg-COD/L) affects the growth and maturation of aquatic animals while a high concentration of methanol e.g., 1,527.60 mg/L (2,291 mg-COD/L) can induce acute toxic effects in fish. Moreover, the high carbon addition in the system may induce the ammonia reproduction from the dissimilatory nitrate reduction to ammonium or DNRA process [22]. Therefore, we can conclude from all the results that

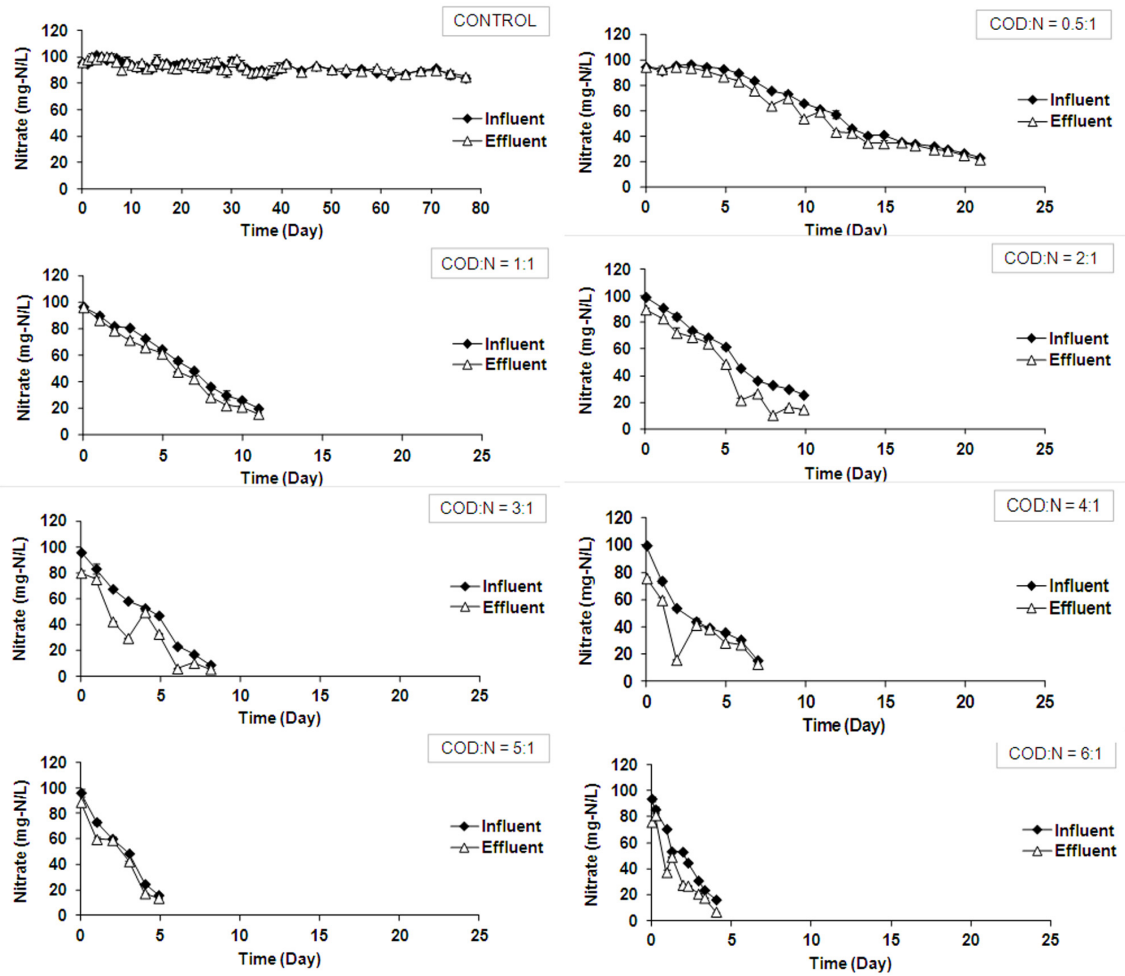


Fig. 6. The alteration of nitrate concentration in the influent and effluent of each COD:N ratio.

Table 1. The nitrate Removal Rates and Efficiencies at Different COD:N Ratios

COD:N ratio	Denitrification rate (mg-N/day)	Nitrate removal efficiency (%)	The amount of methanol added (mg COD/day)	COD depletion (%)
No methanol addition	70.27	0.55 ± 1.17	-	-
0.5:1	1,933.10	7.47 ± 6.40	16.72	6.25
1:1	3,503.09	12.32 ± 8.17	29.2	26.82
2:1	3,806.93	27.27 ± 21.29	26.18	17.44
3:1	5,102.88	33.32 ± 21.18	39.46	-27.97
4:1	4,966.56	20.97 ± 21.29	41.84	-38.08
5:1	7,785.60	13.96 ± 9.42	59.78	-63.03
6:1	9,182.88	31.44 ± 18.44	81.63	-63.38

the proper COD:N ratio which promote the best denitrification activity in a solid digestion column was 3:1. The acceptable amount of excess methanol (as COD) was detected in the outlet water of the digestion column for this COD:N ratio. In comparison, this ratio was less than the number of 3.7:1 from stoichiometric reaction of nitrate removal using methanol as a carbon source [23]. It might be because a portion of the carbon source for the denitrification came from the digestion of the suspended solid retaining in the solid digestion column. Hence, the amount of external carbon source required could be reduced.

4. Conclusions

A single-reactor solid digestion-denitrification column can provide two successive performances of the suspended solid separation and nitrate removal. In the first step of solid removal from aquaculture pond using the simple gravity sedimentation column, the slowest upflow velocity of 0.57 m/h (11.25 L/h) gave the highest solid removal efficiency of $72.72\% \pm 8.24\%$. However, the total suspended solids removal was highest at 18.13 m/h (360 L/h). As a result, the upflow velocity of 18.13 m/h was therefore selected and applied to the solid separation purpose. For denitrification, the digestion of the suspended solid retaining in the solid digestion column was promoted under anaerobic condition. The optimum hydraulic retention time of 1 hour in a reactor by water from the equalization tank was operated, this duration was sufficient for the complete denitrification activity in a solid digestion column. The external carbon source addition (methanol) was essential for this process. The COD:N ratio, which promotes the best denitrification activity in the solid digestion column was 3:1. This was less than the stoichiometric number of 3.7:1 for the nitrate removal using methanol as a carbon source.

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